absolute neutrophil count of ≥500 cells/mm^3 with >75% donor chimerism (RFLP confirmed). The dominating CBU was defined by chimerism >75%. ALDH^{br} dosing was defined as high if the thawed CBU delivered >47,000 ALDH^{br} cells/kg which was identified as predictive of engraftment in single CBT in prior studies.
Results: The median patient age was 28.8 years (range, 3.7-64.8 years) and weight of 68.70 kg (range, 15.2-111.8 kg), 17 male, 13 CMV+. The median TNC per CBU was 2.2×10^7/kg (range, 1.2-10.3×10^7/kg) with the median combined TNC of 3.9×10^7/kg (range, 2.7-18.0×10^7/kg). 5 patients received two high ALDH^{br} units, 10 patients received one high and one low ALDH^{br} unit, and 12 received two low ALDH^{br} units. 23 patients were evaluable for engraftment. 10 of 12 patients receiving ≥1 high ALDH^{br} CBUs engrafted. The other 2 patients died of infection without engraftment (days 29 and 34 post-CBT). Conversely, 4 of 9 patients receiving low ALDH^{br} CBUs failed to engraft. In the high/low group, only the high ALDH^{br} CBUs engrafted. The sensitivity and specificity for ALDH^{br} dose is 0.71 and 0.67, respectively. The positive and negative predictive values are 0.86 and 0.44, respectively. Other graft parameters did not predict the dominating CBU.
Conclusion: Post-thaw ALDH^{br} dosing has a high positive predictive value for predicting engraftment in dCBT. Further studies are planned.

Table 1. ALDHbr Content of Units and Engraftment Status

	ALDHbr Content of Units		
	Low/Low	High/Low	High/High
Total Patients	12	10	5
Evaluable Patients	9	9	5
Engrafted	5	7 [all with high unit]	5
Non-Engrafted	4	2	

297

COMPARATIVE EFFECTIVENESS ANALYSIS OF CD34 + SELECTED, T-CELL DEPLETED (TCD) HLA-MATCHED SIBLING GRAFTS ON ALLOGENEIC HEMATOPOIETIC CELL TRANSPLANTATION FOR PATIENTS WITH ACUTE MYELOID LEUKEMIA (AML) IN COMPLETE REMISSION

Pasquini, M.C.[1], Devine, S.[2], Mendizabal, A.[3], Baden, L.R.[4], Wingard, J.R.[5], Lazarus, H.M.[6], Appelbaum, F.R.[7], Keever-Taylor, C.[1], O'Reilly, R.J.[8,9], Soiffer, R.J.[4] [1] Medical College of Wisconsin, Milwaukee, WI; [2] Ohio State University Medical Center, Columbus, OH; [3] The EMMES Corporation, Rockville, MD; [4] Dana-Farber Cancer Institute, Boston, MA; [5] University of Florida, Shands Cancer Center, Gainesville, FL; [6] Case Western Reserve University, School of Medicine, Cleveland, OH; [7] Fred Hutchinson Cancer Research Center, Seattle, WA; [8] Memorial Sloan-Kettering Cancer Center, New York, NY; [9] On behalf of the Blood and Marrow Transplant Clinical Trials Network

Numerous single institution studies have demonstrated that TCD significantly reduces the incidence of graft-versus-host-disease (GVHD). However, concerns about leukemia relapse, graft rejection, and variability in technique have limited the widespread application of this approach. Promising results of TCD in patients with AML prompted the Blood and Marrow Transplant Clinical Trials Network (BMT CTN) to study a single uniform technique of CD 34+ selection using the Miltenyi CliniMacs device in a Phase II clinical trial (BMT CTN 0303) of adult AML patients in first or second complete remission (CR1/CR2) receiving peripheral blood stem cell (PBSC), HLA-matched sibling donor transplants. We compared outcomes of the 44 patients transplanted on the BMT CTN 0303 trial to a contemporary cohort of 102 patients with AML enrolled on the BMT CTN 0101 – Phase III clinical trial of anti-fungal prophylaxis after myeloablative HCT with pharmacologic immune suppression post-transplant (IST). This analysis compared TCD (BMT CTN #0303) versus IST (BMT CTN 0101) with respect to the endpoints of: neutrophil engraftment (ENG), rates of acute and chronic GVHD, transplant-related mortality (TRM), relapse, disease-free survival (DFS), and overall survival (OS). Groups were similar for patient-, disease- and transplant specific characteristics except for the proportion of patients in CR2 (TCD 7% vs. IST 27%), unfavorable risk cytogenetics (TCD 32% vs. IST 18%), the use of mobilized PBSC (TCD 100% vs. IST 81%) and graft composition (TCD ≤1 x10^5 CD3+ cells/kg). The results revealed lower rates of grades II-IV acute (p = 0.046) and chronic GVHD (p = 0.01) in the TCD group with no difference in ENG, leukemia relapse and TRM. DFS and OS were similar between the two groups in the univariate setting and also after adjustment for potential prognostic factors using a Cox proportional hazards model. Reduction of GVHD rates without an increase in relapse rates and no requirement for post-transplant immunosuppression are distinct advantages of this method of TCD. These results support the extension of this approach to the unrelated donor setting and additional larger, prospective studies to definitively address the role of rigorous TCD in HCT.

TCD vs IST Outcomes

Outcome	TCD % (95% Confidence Interval) N = 44	IST % (95% Confidence Interval) N = 102
DFS at 6 mo	81 (66-90)	75 (68-87)
Relapse at 12 mo	19 (6-32)	19 (11-26)
TRM at 12 mo	19 (6-31)	22 (14-30)
ENG at 28 d	100 (86-100)	90 (79-100)
Acute GVHD II-IV at 100 d	20 (9-32)	37 (28-47)
Acute GVHD III-IV at 100 d	5 (0-11)	10 (4-16)
Chronic GVHD at 12 mo	19 (7-32)	47 (35-58)
Overall Survival at 12 mo	74 (57-85)	69 (59-77)

298

PRECLINICAL EVALUATION OF HUMAN NOR-EPINEPHRINE TRANSPORTER (HNET)/MIBG REPORTER SYSTEM FOR IMAGING ADOPTIVELY TRANSFERRED EBV-SPECIFIC CYTOTOXIC T-LYMPHOCITES

Doubrovin, M., Doubrovina, E., Moroz, M., Pankov, D., Wang, X., Olszewska, M., Riviere, I., O'Reilly, R., Larson, S., Blasberg, R. Memorial Sloan-Kettering Cancer Center, New York, NY

Background: We previously demonstrated hNET/MIBG to be a reporter gene system feasible for non-invasive quantitative monitoring of cytotoxic T-cells (CTLs) in vivo. In this preclinical study, we test whether the hNET reporter gene construct, under conditions simulating adoptive immune cell therapy in patients, is potentially applicable imaging paradigm.
Methods: The hNET reporter gene was cloned into a clinical grade SFG pseudotyped MoMLV retroviral backbone, packaged in PG-13 retroviral producer cell line and was used to transfect EBV-specific T cells. Following the future clinical protocol, EBV-specific T cells were pre-generated from a normal donor by stimulation with autologous EBV-transformed B-cells, as we previously described. Based on expression of LNGFR, a selection gene in the same retroviral cassette, reporter-gene expressing cells (CTL-NIN) were selected by FACS sorting, characterized for cytotoxicity and frozen for long-term storage. Initially, ^{123}I-MIBG uptake was evaluated in the CTL-NIN retrieved from liquid nitrogen storage according to GMP requirements. In the preclinical study, upon thawing, prior to adoptive transfer, CTLs were preincubated with 100 microCi/ml (3.73 MBq/ml) of ^{124}I-MIBG for 2 hours and injected into the EBV-BLCL xenograft tumor model bearing NOD-SCID mice. Assessment MIBG radioactivity in CTL-NIN prior to injection and MicroPET imaging of the injected radiolabeled cells in a phantom and in the animals, with post-mortem ex-vivo radioactivity measurements were performed.
Results: CTL-NINs were produced according to GMP procedures appeared to have >90% reporter gene expression post-sorting with improved specificity to EBV-BLCL targets (35% with 10:1 E:T ratio). Storage and thawing decreased their cytotoxicity (25%) and ^{123}I-MIBG uptake (217 ml/g, compared to 330 ml/g pre-freezing). ^{124}I-MIBG labeling of CTL-NIN T cells ex-vivo was sufficient for in vivo imaging. In vivo microPET imaging confirmed our ability to detect 10^5 ^{124}I-MIBG ex vivo prelabeled CTL-NIN distributed in a 1 cm^3 tumor volume.
Conclusions: 1) hNET transduced CTLs produced and prepared for adoptive immune cell transfer according to a clinical protocol